PATENT COOPERATION TREATY

PCT

12

REC'D 11 JAN 2002
WIPO PCT

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference	FOR FURTHER ACTION	See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416)
05213-0551 WP		
International application No.	International filing date *(day/month/year)*	Priority date *(day/month/year)*
PCT/US00/25166	14/09/2000	14/09/1999
International Patent Classification (IPC) or national classification and IPC C12N15/12		
Applicant ENTREMED, INC. et al.		

1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.

2. This REPORT consists of a total of 7 sheets, including this cover sheet.

   ☐ This report is also accompanied by ANNEXES, i.e. sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

   These annexes consist of a total of sheets.

3. This report contains indications relating to the following items:

I	☒	Basis of the report
II	☒	Priority
III	☐	Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
IV	☒	Lack of unity of invention
V	☒	Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations suporting such statement
VI	☐	Certain documents cited
VII	☒	Certain defects in the international application
VIII	☐	Certain observations on the international application

Date of submission of the demand	Date of completion of this report
16/04/2001	08.01.2002
Name and mailing address of the international preliminary examining authority: European Patent Office D-80298 Munich Tel. +49 89 2399 - 0 Tx: 523656 epmu d Fax: +49 89 2399 - 4465	Authorized officer Nichogiannopoulou, A Telephone No. +49 89 2399 8054

Form PCT/IPEA/409 (cover sheet) (January 1994)

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No. PCT/US00/25166

I. Basis of the report

1. With regard to the **elements** of the international application *(Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report since they do not contain amendments (Rules 70.16 and 70.17))*:
Description, pages:

1-56 as originally filed

Claims, No.:

1-23 as originally filed

Drawings, sheets:

1/5-5/5 as originally filed

Sequence listing part of the description, pages:

1-8, filed with the letter of 19.12.2000

2. With regard to the **language**, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.

These elements were available or furnished to this Authority in the following language: , which is:

- ☐ the language of a translation furnished for the purposes of the international search (under Rule 23.1(b)).
- ☐ the language of publication of the international application (under Rule 48.3(b)).
- ☐ the language of a translation furnished for the purposes of international preliminary examination (under Rule 55.2 and/or 55.3).

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

- ☐ contained in the international application in written form.
- ☐ filed together with the international application in computer readable form.
- ☒ furnished subsequently to this Authority in written form.
- ☒ furnished subsequently to this Authority in computer readable form.
- ☒ The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.
- ☒ The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

4. The amendments have resulted in the cancellation of:

- ☐ the description, pages:
- ☐ the claims, Nos.:
- ☐ the drawings, sheets:

5. ☐ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed (Rule 70.2(c)):

   (Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report.)

6. Additional observations, if necessary:

II. Priority

1. ☐ This report has been established as if no priority had been claimed due to the failure to furnish within the prescribed time limit the requested:

   ☐ copy of the earlier application whose priority has been claimed.

   ☐ translation of the earlier application whose priority has been claimed.

2. ☐ This report has been established as if no priority had been claimed due to the fact that the priority claim has been found invalid.

   Thus for the purposes of this report, the international filing date indicated above is considered to be the relevant date.

3. Additional observations, if necessary:
   see separate sheet

IV. Lack of unity of invention

1. In response to the invitation to restrict or pay additional fees the applicant has:

   ☐ restricted the claims.

   ☐ paid additional fees.

   ☐ paid additional fees under protest.

   ☐ neither restricted nor paid additional fees.

2. ☐ This Authority found that the requirement of unity of invention is not complied and chose, according to Rule 68.1, not to invite the applicant to restrict or pay additional fees.

3. This Authority considers that the requirement of unity of invention in accordance with Rules 13.1, 13.2 and 13.3 is

   ☐ complied with.

☐ not complied with for the following reasons:

4. Consequently, the following parts of the international application were the subject of international preliminary examination in establishing this report:

☒ all parts.

☐ the parts relating to claims Nos. .

V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Yes:	Claims	11-23
	No:	Claims	1-10
Inventive step (IS)	Yes:	Claims	11-23
	No:	Claims	1-10
Industrial applicability (IA)	Yes:	Claims	1-23
	No:	Claims	

2. Citations and explanations
see separate sheet

VII. Certain defects in the international application

The following defects in the form or contents of the international application have been noted:
see separate sheet

Re Item II
Priority

1. The present application validly claims priority from 14.09.1999. Any documents cited in the International Search Report as P documents have therefore not been considered as comprised in the prior art relevant for the present application.

Re Item IV
Lack of unity of invention

1. The present application relates to processes for the recombinant production (claims 1-10) and purification (claims 11-23) of Endostatin™. Since both Endostatin™ as well as its recombinant production are well known from the prior art (see D below), there is no special technical feature, i.e. a contribution each of these processes makes over the prior art, unifying them into a single invention. The IPEA therefore agrees with the objection put forward by the ISA as to lack of unity pursuant to Rule 13 PCT, and considers that the present application relates to two distinct groups of inventions. However, since all claims could be searched without effort justifying additional fees, the two inventions will be examined jointly without requirement of additional examination fees. Should the application enter the European phase, an objection under the corresponding EPC article will be raised.

Re Item V

Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Reference is made to the following documents:

   D1: WO 99 26480 A (GENETIX PHARMACEUTICALS INC; MASSACHUSETTS INST TECHNOLOGY (US)) 3 June 1999 (1999-06-03)

   D2: JOHN H ET AL: 'Novel glycosylated forms of human plasma endostatin and circulating endostatin-related fragments of collagen XV.' BIOCHEMISTRY, vol. 38, no. 32, 10 August 1999 (1999-08-10), pages 10217-10224, XP002162137

   D3: DHANABAL M ET AL: 'Endostatin: Yeast production, mutants, and antitumor effect in renal cell carcinoma' CANCER RESEARCH, vol. 59, no. 1, 1999, pages 189-197, XP002100110

   D4: BOEHM T ET AL: 'Zinc-binding of endostatin is essential for its antiangiogenic activity' BIOCHEMICAL AND BIOPHYSICAL RESEARCH COMMUNICATIONS, vol. 252, 1998, pages 190-194, XP002100108

   D5: BOEHM T ET AL: 'Disruption of the KEX1 gene in Pichia pastoris allows expression of full-length murine and human endostatin' YEAST, vol. 15, 1999, pages 563-572, XP002109421

2. **Novelty and Inventive step** (Article 33(2) and (3) PCT)
   The present application discloses methods for the recombinant production of Endostatin™ (claims 1-10) as well as methods for purifying Endostatin™ (claims 11-23), leading to large scale recovery of the protein.

2.1. **D1** discloses the recombinant expression of human Endostatin in cells transduced with constructs having 100% identity with SEQ ID Nos:3, 4, 5, 6, 8 and 11 (Examples 1-4). **D1** is thus detrimental to the novelty and inventive step of claims 1 and 5-7.

2.2. **D2** discloses the chromatographic isolation of Endostatin proteins from human blood ultrafiltrate through cation exchange and RP chromatography and the subsequent

lyophilisation of aliquots. Recombinant endostatin can be produced by e.g. *Pichia pastoris*, *E. coli*, baculovirus or human embryonic kidney cells. **D2** is thus detrimental to the novelty and inventive step of claims 1-8.

2.3. **D3-D5** disclose the recombinant production of murine Endostatin in *Pichia pastoris* and its subsequent purification. **D3-D5** are thus detrimental to the novelty and/or inventive step of claims 1 and 3-10.

3. **Industrial applicability** (Article 33(4) PCT)
The subject-matter of the present set of claims appears to be industrially applicable under the terms of Article 33(4) PCT.

Re Item VII

Certain defects in the international application

1. Contrary to the requirements of Rule 5.1(ii) PCT, documents **D1-D5** are not identified in the description and the relevant background art disclosed therein is not briefly discussed.